

We claim:

1. A process of making a human-like glycoprotein in a lower eukaryotic host cell comprising the step of introducing into the cell an *N*-acetylglucosaminyltransferase III activity.
2. A process of making a human-like glycoprotein in a lower eukaryotic host cell comprising the step of expressing in the cell an *N*-acetylglucosaminyltransferase III activity.
3. A process of making a human-like glycoprotein in a lower eukaryotic host cell comprising the step of expressing in the cell one or more enzymatic activities that produce *N*-glycans comprising GlcNAc<sub>3</sub>Man<sub>3</sub>GlcNAc<sub>2</sub>, GlcNAc<sub>2</sub>Man<sub>3</sub>GlcNAc<sub>2</sub> or GlcNAc<sub>2</sub>Man<sub>5</sub>GlcNAc<sub>2</sub> bisected structures.
4. The process of claims 1 or 2, wherein the *N*-acetylglucosaminyltransferase III activity produces a bisected glycan.
5. The process of claims 1 or 2, wherein the glycoprotein comprises a bisected glycan.
6. The process of claims 1 or 2, wherein the activity is substantially intracellular.
7. The process of claims 1, 2, or 3, further comprising the step of isolating the glycoprotein from the host cell.
8. The process of claims 1, 2, or 3, wherein the host cell is selected from the group consisting of *Pichia pastoris*, *Pichia finlandica*, *Pichia trehalophila*, *Pichia koclamae*, *Pichia membranaefaciens*, *Pichia opuntiae*, *Pichia thermotolerans*, *Pichia salictaria*, *Pichia guercuum*, *Pichia pijperi*, *Pichia stiptis*, *Pichia methanolica*, *Pichia* sp., *Saccharomyces cerevisiae*, *Saccharomyces* sp., *Hansenula polymorpha*, *Kluyveromyces* sp., *Candida albicans*, *Aspergillus nidulans*, *Aspergillus niger*, *Aspergillus oryzae*, *Trichoderma reesei*, *Chrysosporium lucknowense*, *Fusarium* sp., *Fusarium gramineum*, *Fusarium venenatum*, and *Neurospora crassa*.

9. The process of claim 8, wherein the host cell is selected from the group consisting of *Pichia pastoris*, *Pichia finlandica*, *Pichia trehalophila*, *Pichia koclamae*, *Pichia membranaefaciens*, *Pichia opuntiae*, *Pichia thermotolerans*, *Pichia salictaria*, *Pichia guercuum*, *Pichia pijperi*, *Pichia stiptis*, *Pichia methanolica*, and *Pichia* sp..

10. The process of claim 9, wherein the host cell is *Pichia pastoris*.

11. The process of claims 1, 2, or 3, wherein the glycoprotein is a therapeutic protein.

12. The process of claim 11, wherein the therapeutic protein is selected from the group consisting of erythropoietin, cytokines, coagulation factors, soluble IgE receptor  $\alpha$ -chain, IgG, IgG fragments, IgM, interleukins, urokinase, chymase, urea trypsin inhibitor, IGF-binding protein, epidermal growth factor, growth hormone-releasing factor, annexin V fusion protein, angiostatin, vascular endothelial growth factor-2, myeloid progenitor inhibitory factor-1, osteoprotegerin,  $\alpha$ -1-antitrypsin,  $\alpha$ -feto protein, and DNase II.

13. A lower eukaryotic host cell comprising an *N*-acetylglucosaminyltransferase III activity.

14. A lower eukaryotic host cell comprising an *N*-acetylglucosaminyltransferase II activity and an *N*-acetylglucosaminyltransferase III activity.

15. The host cell of claim 13 or 14, wherein the activity is substantially intracellular.

16. The host cell of claim 13 or 14, wherein the cell produces *N*-glycans comprising GlcNAcMan<sub>3</sub>GlcNAc<sub>2</sub> structures that are capable of reacting with GnTIII activity.

17. The host cell of claim 13 or 14, wherein the *N*-acetylglucosaminyltransferase III activity produces a bisected glycan.

18. A lower eukaryotic host cell comprising an *N*-glycan that comprises a bisected glycan.

19. The host cell of claim 18, wherein the *N*-glycan contains greater than 10 mole % of the bisected glycan.

20. The host cell of claim 18, wherein the *N*-glycan contains greater than 80 mole % of the bisected glycan.

21. A lower eukaryotic host cell comprising an *N*-glycan that comprises GlcNAc<sub>3</sub>Man<sub>3</sub>GlcNAc<sub>2</sub>, GlcNAc<sub>2</sub>Man<sub>3</sub>GlcNAc<sub>2</sub> or GlcNAc<sub>2</sub>Man<sub>5</sub>GlcNAc<sub>2</sub> bisected structures.

22. The host cell of claims 13, 14, 18, or 21, wherein the host cell is selected from the group consisting of *Pichia pastoris*, *Pichia finlandica*, *Pichia trehalophila*, *Pichia koclamae*, *Pichia membranaefaciens*, *Pichia opuntiae*, *Pichia thermotolerans*, *Pichia salictaria*, *Pichia guercuum*, *Pichia pijperi*, *Pichia stiptis*, *Pichia methanolica*, *Pichia* sp., *Saccharomyces cerevisiae*, *Saccharomyces* sp., *Hansenula polymorpha*, *Kluyveromyces* sp., *Candida albicans*, *Aspergillus nidulans*, *Aspergillus niger*, *Aspergillus oryzae*, *Trichoderma reesei*, *Chrysosporium lucknowense*, *Fusarium* sp., *Fusarium gramineum*, *Fusarium venenatum*, and *Neurospora crassa*.

23. The host cell of claim 22, wherein the host cell is selected from the group consisting of *Pichia pastoris*, *Pichia finlandica*, *Pichia trehalophila*, *Pichia koclamae*, *Pichia membranaefaciens*, *Pichia opuntiae*, *Pichia thermotolerans*, *Pichia salictaria*, *Pichia guercuum*, *Pichia pijperi*, *Pichia stiptis*, *Pichia methanolica*, and *Pichia* sp..

24. The host cell of claim 23, wherein the host cell is *Pichia pastoris*.

25. A lower eukaryotic host cell comprising a Man<sub>5</sub>GlcNAc<sub>2</sub> core structure or a Man<sub>3</sub>GlcNAc<sub>2</sub> core structure modified by a bisecting GlcNAc.

26. The host cell of claim 25, wherein the cell produces greater than 10 mole % of the modified structure.

27. The host cell of claim 25, wherein the cell produces greater than 50 mole % of the modified structure.

28. A lower eukaryotic host cell comprising an *N*-acetylglucosaminyltransferase I activity and an *N*-acetylglucosaminyltransferase III activity.

29. The host cell of claim 28, wherein the activities are substantially intracellular.

30. The host cell of claim 28, wherein the cell produces *N*-glycans comprising GlcNAcMan<sub>3</sub>GlcNAc<sub>2</sub> that are capable of reacting with GnTIII activity.

31. The host cell of claim 28, wherein the *N*-acetylglucosaminyltransferase III activity produces a bisected glycan.

32. A lower eukaryotic host cell comprising a GnTIII activity and a mannosidase II activity.

33. The host cell of claim 32, further comprising an *N*-acetylglucosaminyltransferase I activity.

34. The host cell of claim 32, further comprising an *N*-acetylglucosaminyltransferase II activity.

35. The host cell of claim 32, further comprising an *N*-acetylglucosaminyltransferase I activity and an *N*-acetylglucosaminyltransferase II activity.

36. The host cell of claims 13, 14, 28, or 32 that is deficient in an *OCH1* mannosyltransferase activity.

37. The host cell of claims 13, 14, 28, or 32 that is deficient in a Dol-P-Man:Man<sub>5</sub>GlcNAc<sub>2</sub>-PP-Dol mannosyltransferase activity.

38. The host cell of claims 13, 14, 28, or 32, further comprising an  $\alpha$ -1,2-mannosidase I activity.

39. The host cell of claims 13, 14, 28, or 32, further comprising a UDP-GlcNAc transporter.

40. The glycoprotein made by the process of claim 1.

41. The glycoprotein made by the process of claim 2.

42. The glycoprotein made by the process of claim 3.

43. The glycoprotein made by the process of claim 8.

44. The glycoprotein made by the process of claim 9.

45. The glycoprotein made by the process of claim 10.

46. The glycoprotein made by the process of claim 11.

47. The glycoprotein made by the process of claim 12.

48. A glycoprotein comprising a bisecting GlcNAc on a Man<sub>5</sub>GlcNAc<sub>2</sub> or a Man<sub>3</sub>GlcNAc<sub>2</sub> core structure produced in a lower eukaryotic host cell.

49. A glycoprotein comprising a bisecting GlcNAc attached to a Man<sub>5</sub>GlcNAc<sub>2</sub>, Man<sub>4</sub>GlcNAc<sub>2</sub>, Man<sub>3</sub>GlcNAc<sub>2</sub>, GlcNAcMan<sub>3</sub>GlcNAc<sub>2</sub>, GlcNAcMan<sub>5</sub>GlcNAc<sub>2</sub>, or a GlcNAc<sub>2</sub>Man<sub>3</sub>GlcNAc<sub>2</sub> core structure produced in a lower eukaryotic host cell.

50. The glycoprotein of claim 49, wherein greater than 10 mole % of the core structures are modified by the bisecting GlcNAc.

51. The glycoprotein of claim 49, wherein greater than 80 mole % of the core structures are modified by the bisecting GlcNAc.

52. A pharmaceutical composition comprising the glycoprotein of any one of claims 40 - 51.

53. A vector selected from the group consisting of pVA, pVB, and pVC.

54. The vector of claim 53, wherein the vector is selected from the group consisting of pVA53, pVA54, pVA55, pVB53, pVB54, and pVB55.